ISSN 0377-9335

ENTOMON

Vol. 39 September 2014 No. 3

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ASSOCIATION FOR ADVANCEMENT OF ENTOMOLOGY
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ENTOMON

ENTOMON is a quarterly journal published by the Association for Advancement of Entomology devoted to the publication of research work on various aspects of insects and related branches of Entomology.

Address all MS and editorial correspondence to the Chief Editor, ENTOMON, Department of Entomology, College of Agriculture, Kerala Agricultural University, Vellayani, Thiruvananthapuram 695 522, Kerala, India. E mail: ***editor.entomon@kau.in***

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ENTOMON 39(3): 119-128 (2014)
Article No. ent. 39301

Infestation induced biochemical reactions on papaya by mealybug, *Paracoccus marginatus* Williams and Granara de Willink (Hemiptera: Pseudococcidae)

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ABSTRACT: *Paracoccus marginatus* Williams and Granara de Willink, (Hemiptera: Pseudococcidae) is an invasive alien sucking pest that attacks several genera of economically important tropical and subtropical plants. Biochemical study revealed that total protein content was very low (1.88mg/g) in plants inoculated with third instar nymphs of papaya mealybug, whereas the plants inoculated with adult mealybugs recorded a higher protein concentration of 4.60 mg/g. Similarly, plants inoculated with third instar nymph of papaya mealybug had a decreased mean concentration (0.019 mg/g) of Indole Acetic Acid (IAA) whereas plant inoculated with adult mealybugs recorded the highest mean IAA concentration was 0.070 mg/g. The papaya seedlings uninfested with papaya mealybug recorded the highest protein (5.34 mg/g) and IAA (0.185 mg/g) content. However, the gibberellic acid content of 5μg/g was estimated from the leaves of plants showing crinkling symptom infested with third instar nymphs and the uninfested papaya seedlings recorded lower levels of gibberellic acid content of 1 μg/g.

KEYWORDS: Papaya mealybug, *Paracoccus marginatus*, total protein, Indole-3-Acetic Acid, Gibberellic Acid.

INTRODUCTION

Papaya mealybug, *Paracoccus marginatus* Williams and Granara de Willink, native to Mexico and Central America (Miller *et al*., 1999), is an invasive insect pest has been first reported in India from Coimbatore (Tamil Nadu, India) on papaya, jatropha and certain other plants in 2008 (Muniappan *et al*., 2008, Regupathy and Ayyasamy, 2011). Later it was recorded in Kerala (Krishnakumar and Rajan, 2009; Lyla and Philip, 2010), Karnataka, Andhra Pradesh, Maharastra, Tripura and Odisha. Approximately 95 host plant species belonging to 39 families

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were recorded to be infested by *P. marginatus* in Kerala (Manichellappan *et al*., 2013).

Immature and adult stages of *P. marginatus* suck the sap by inserting their stylets into the epidermis of the leaf resulting in curling, crinkling, rosetting, twisting and leaf distortion. The honey dew excreted by the mealybugs and the associated black sooty mould formation impairs photosynthetic efficiency of the infested plants. Flowers fail to open and petals become twisted or malformed. Fruits may be unusually small and such fruits eventually shrivel and drop. Premature flower drop and poor fruit set occur, subsequently.

Injuries to plants by insects result in different physiological and biochemical responses than mechanical damage alone. Generally, in plants the endogenous growth hormone, auxin is synthesized in young expanding leaves at the shoot apex and is actively transported down to the plant. Saikia *et al.* (2011) observed that the attack of *Helopeltis theivora* Waterhouse on the axillary vegetative buds and young leaves of tea resulted in decreased level of auxin than non infested plants. However, very little is understood about the reactions in host plants due to the *P. marginatus* infestation.

MATERIALS AND METHODS

Mass culturing of *P. marginatus* was done on sprouted potatoes as standardised by Gautam (2008). Three months old healthy papaya seedlings with an average height of 15cm with 4-5 leaves were selected and ensured the seedlings free from pests, diseases and nutrient deficiency. In case of protein and IAA content estimation, total of 33 seedlings were replicated thrice at the rate of 11 seedlings per replication. Single 1st instar nymph (crawler) of papaya mealybug was transferred to group I, two crawlers to group II, three crawlers to group III and so on upto ten crawler to group X. Similarly 2nd, 3rd instar nymphs and adult mealy bug were also transferred to papaya seedlings. Uninfested papaya seedlings were maintained as untreated control. In case of GA estimation, papaya seedlings infested with five 3rd instar nymph of papaya mealy bug and unifested plants were analysed without any change in the number/ stage of insect, owing to the large sample size required for the analysis.

The healthy and infested leaves from the growing point were collected individually and subjected to the analyses of total protein and IAA.

Total protein

Leaf samples (500mg) were taken from the growing point of plant and were groundedwell in 10ml cold distilled water using mortar and pestle. The ground sample was spun at 5000 rpm for 10 minutes in a refrigerated centrifuge (REMI®, CFC free, C-24). The supernatant was collected and 0.1ml aliquot was taken for analysis. Total protein content in healthy and infested leaves of papaya was estimated as per Lowry *et al*. (1951).

Bovine serum albumin (BSA) was used as standard. Folin – phenol reagent (ready mix; 0.5 ml) was added to the sample tubes and incubated at room temperature for 30 minutes. After

incubation, absorbance was measured at 660nm by UV spectrophotometer (ELICO®). A standard graph was drawn by plotting concentration (mg) of protein on the X-axis and absorbance (nm) on the Y-axis. From the graph total protein content in the sample was estimated and expressed as mg/g plant tissue.

Indole Acetic Acid (IAA)

IAA present in healthy and infested leaves of papaya were estimated using spectrophotometric method standardised to suit the present investigation.

Indole acetic acid (10 mg) weighed and dissolved in 100 ml of 0.1M Na_2CO_3 to prepare 100 ppm stock. From the stock solution working standards were prepared *viz*., 10, 20, 30, 40, 50, 60 ppm. The absorbance was measured at 540 nm in spectrophotometer. A standard graph was drawn by plotting concentrations of IAA (mg) on X-axis and absorbance (nm) on the Y-axis.

Leaf sample (500 mg) was macerated with 10ml cold distilled water. The content was spun at 5000 rpm (10 minutes) in a refrigerated centrifuge. The supernatant was collected, filtered and the volume was made up to 25 ml with ice cold distilled water. Two sets of 1ml of aliquot in a test tube was taken and 1ml each of phosphate buffer (68 ml of 0.2 M monobasic-NaH_2PO_4 + 32 ml of 0.2M dibasic-Na_2HPO_4 and made up to 200 ml with cold distilled water) and distilled water was added. Distilled water alone (2 ml) served as blank and to which 1ml of phosphate buffer was added. To stop the reaction, the first set of tubes (control) was kept in hot water bath for 10-20 seconds. The content was then cooled and 8 ml of Garden Webber reagent (mix 2 ml of 0.5M ferric chloride +100 ml of 35% perchloric acid) was added. Pink colour developed and the absorbance was measured at 540 nm. Simultaneously, second set of tubes were kept at room temperature for 1h. After 1h the test tubes were placed in hot water bath (10-20 seconds) to stop the reaction. The content was cooled and Garden Webber reagent (8ml) was added. Pink colour developed was measured at 540 nm in the spectrophotometer. The absorbance value was plotted in the standard graph and the corresponding concentrations (X μg) were recorded. Using the concentration obtained from graph, IAA content was calculated and expressed as mg of unoxidised auxin per gram of plant sample.

Gibberellic acid (GA)

The method of extraction and purification of endogenous level of gibberellic acid (GA) in plant samples was extensively modified from those described by House (1961). Gibberellic acid present in the plants was estimated based on the conversion to gibberellic acid followed by the measurement of its absorption at 254 nm. As the procedure involved large quantity of the growing tissues, infested plant parts that exhibit greater extent of damage due to third instar nymph were used.

Extraction of free gibberellins from plants

Gibberellins occur in plants in bound and free form. The free gibberellins from the plant

samples were extracted by the following procedure.

Fresh plant sample (2g) was homogenized with 20 ml chilled methanol (80% v/v) and left overnight at 4°C. The extract was filtered through Whatman No. 40 filter paper and solid residue further isolated by centrifugation at 10000 rpm for 5 minutes with methanol. The methanolic extracts are pooled and concentrated to a water residue in vacuum (30-40°C) by rotary evaporator (Superfit®). The volume was adjusted to 10 ml with 0.2M PO_4 buffer (pH 7.5). The methanol compounds were removed by partitioning it twice with 5 ml methyl ether in a 20 ml glass vial. The ether was layered to aqueous phase and two phases system was gently stirred for three minutes in a magnetic stirrer. After discarding the ether phase, the aqueous phase was adjusted to pH 2.7 with 1M HCl. The aqueous phase partitioned thrice against 10 ml of ethyl acetate and the ethyl acetate layer was further partitioned twice against 0.4M $NaHCO_3$. The aqueous phase was adjusted to pH 2.5 with 1.6M HCl. The acidified phase was partitioned two times against 10 ml ethyl acetate. The ethyl acetate layer was dissolved in methanol and stirred in vials at 4°C.

Sample (1.5 ml) containing GA extracted as per the above was pipetted out to the test tube and 2 ml of zinc acetate was added. After 2 minutes, 2 ml of potassium ferrocyanide was added and centrifuged at low speed (3000rpm) for 15 minutes. From this, 5 ml of supernatant was taken and 5 ml of HCl (30%) was added and incubated the mixture at 20°C for 75 minutes. The blank sample was treated with HCl (5%) and the absorbance of the sample and the blank was measured at 254 nm. The sample absorbance was plotted in the standard graph and the corresponding concentrations (X µg) were recorded for calculation of GA which was expressed as µg per gram of plant tissue.

Analysis of data

Protein, IAA and GA concentrations in plants under each experiment were tabulated and analysed statistically by one sample t-test using the statistical package, SPSS Version 16.

RESULTS AND DISCUSSION

Mealy bug infestation caused a marked variation in the total protein, indole acetic acid (IAA) and giberellic acid (GA) content of the host plants.

Total protein

The concentration of total protein was 4.94 mg/g of leaf sample when the plants were inoculated with one crawler and it was reduced to 3.16 mg/g with the release of ten crawlers per plant. Whereas, a total protein concentration of 4.57 mg/g was estimated from the plants inoculated with one second instar nymph; whereas plants with ten second instar nymphs recorded a protein concentration of 2.73. mg/g. Papaya plants inoculated with third instars showed a decrease in total protein content compared to those plants inoculated with first, second and adult mealybugs. A total protein content of 3.10 mg/g was recorded on plants inoculated with

one third instar nymph of papaya mealybug whereas the protein concentration was reduced to 1.88 mg/g in plants inoculated with ten third instar nymphs. When the plants were inoculated with adult mealybugs there was no noticeable variation was observed in total protein concentration. As the number of mealybugs per plant was increased from one to ten, the total protein concentration decreased from 4.96 to 4.17 mg/g. The result of the experiment showed that the mean concentration of total protein was very low in plants inoculated with third instar nymph (1.88mg/g) of papaya mealybug, whereas the plants inoculated with adult mealybug recorded a maximum protein concentration of 4.66 mg/g (Table 1).

There was a consistent inverse relationship between the level of damage by the insect and protein content in host plants. The lowest level of protein was detected in the leaves infested with ten numbers of third instars of papaya mealybug. As the number of mealybugs increased, there was a decrease in the protein content. Similarly, Eid *et al*. (2011) studied the impact of

Table 1. Effect of infestation of *P. marginatus* on the total protein content of papaya seedling

No. of mealybugs released/plant	Mean protein concentration (mg/g) of leaf sample			
	I instar	II instar	III instar	Adult
1	4.94	4.57	3.10	4.96
2	4.91	4.51	3.02	4.94
3	4.80	4.03	3.01	4.94
4	4.76	3.84	2.96	4.75
5	4.56	3.64	2.93	4.69
6	4.39	3.59	2.94	4.65
7	4.23	3.48	2.86	4.55
8	4.17	3.03	2.56	4.54
9	3.56	2.92	2.49	4.44
10	3.16	2.73	1.88	4.17
Mean	4.35	3.63	2.78	4.66
t value	4.20**	7.50**	20.10**	5.90**
Control	5.34	5.18	5.20	5.14

Mean of 3 replications

** Significant at 1% level

pink mealy bug, *Saccharococcus sacchari* infestation on chemicals and allelo chemicals of some sugar cane cultivars, among them the crude protein content in the infested cane was significantly lower than the un infested cane cultivars. Pitan *et al*. (2011) found that protein, fat, carbohydrate, ash, crude fibre and moisture contents were depleted with increase in mealybug, *Rastrococcus invadens* William population in mango. According to Khattab and Khattab (2005), the total soluble protein of infested leaves of eucalyptus was lower (1.75±0.61) than those of the healthy ones (2.0±0.89 mg/g) due to feeding by gall-forming psyllid. Miles (1999) found that phloem feeding insects established a sustained interaction with sieve elements (SEs). They released saliva that inhibited plant stress responses and prevents closure of pierced SEs by callose or polymerized proteins. Drain of assimilates towards the insect away from other plant parts might contribute to such metabolites reduction (Miles, 1989).

Indole -3 Acetic Acid

With the infestation of one first instar crawler, the IAA content was 0.131 mg/g. It was reduced to 0.022 mg/g when the pest load increased to ten crawlers of first instar on the plants. An estimated IAA content of 0.035 mg/g was recorded on papaya plants inoculated with one second instar nymph and it was reduced to 0.023 mg/g with increase in pest load to ten second instar nymphs per plant. Papaya plants inoculated with third instar nymphs of mealybug resulted a decreased level of IAA content which ranged from 0.022 mg/g and 0.017 mg/g for one and ten crawlers, respectively. When plants were inoculated with one adult mealybug, the IAA content was 0.127 mg/g and it was reduced to 0.070 mg/g with increase in population to ten adults per plant (Table 2).

The analysis showed that the papaya plants inoculated with third instar nymph of papaya mealybug had a reduced mean level of IAA content (0.019 mg/g), followed by plants inoculated with second instar nymphs of mealybug (0.028 mg/g). Whereas estimated level of mean IAA content from the plants inoculated with first instar and adult mealybugs was 0.06 and 0.07 mg/g. The control plants showed higher IAA content of 0.099 to 0.185 mg/g of leaf sample. From the above results, it was inferred that the auxin content recorded on papaya seedlings showed a gradual decrease in their concentration due to *P. marginatus* infestation load.

Infestation by insects brings about a change in level of endogenous growth regulators that result in abnormal development and growth forms. These include epinasty (leaf and stem distortion), hypertrophy (cell enlargement), hyperplasic (cell proliferation), internodal shortening, proliferation of adventitious buds, unusual rooting, flowering or fruiting pattern, abnormal organ development and irregular bud accession (Allen, 1947; Carter, 1973).

Indole 3- acetic acid (IAA) is a naturally occurring auxin which is continuously produced in young meristematic tissues and more rapidly transported to other tissues. Any damage in apical portion can cause a change in auxin synthesis and its concentration in plants. It is evident from the study that the mealybug infestation was concentrated to young shoots which may cause an inhibition in the synthesis of IAA at meristematic region and thereby caused a decrease in IAA content in infested plants.

Table 2. Effect of infestation of *P. marginatus* on the IAA content of papaya seedling

No. of mealybugs released/plant	IAA concentration (mg/g) of leaf sample			
	I instar	II instar	III instar	Adult
1	0.131	0.035	0.022	0.127
2	0.129	0.034	0.022	0.125
3	0.127	0.032	0.021	0.124
4	0.126	0.031	0.019	0.093
5	0.123	0.031	0.019	0.091
6	0.026	0.028	0.019	0.076
7	0.025	0.026	0.019	0.062
8	0.024	0.025	0.018	0.025
9	0.022	0.024	0.017	0.025
10	0.022	0.023	0.017	0.022
Mean	0.060	0.028	0.019	0.070
t value	6.1**	50.1**	55.6**	4.3**
Control	0.169	0.099	0.185	0.135

Mean of 3 replications

** Significant at 1% level

The IAA content in the papaya mealy bug infested leaves was less as compared to uninfested leaves. The IAA content showed a decreasing trend from first group to tenth group in all the stages of infestation. The per cent decrease of IAA which is very high in papaya plants infested with third instars of papaya mealybug. Similarly, aphids substantially reduced the plant growth by their feeding. Honeydew from aphids contains a wide variety of chemicals ingested from the host, including auxins, gibberellins, and cytokinins (Phillips and Cleland 1972; Hussain *et al.* 1973). Marked changes in the normal hormone balance occur within 10 days of infestation by aphids, with growth inhibitors increasing and growth promoters decreasing (Hussain *et al.* 1973). These authors suggested that the large drain on the plant food and hormone resources might be sufficient to account for the growth reduction observed. Saikia *et al*. (2011) found that red spider mite infested plum tree had lower level of auxin than the non-infested one. Bari and Jones (2009) found that blocking of auxin responses had been shown to increase resistance in plants. The tea mosquito bug, *Helopeltis* sp. attack on the axillary vegetative buds and young leaves of tea resulted in decrease in auxin content.

Gibberellic Acid

The gibberellic acid was estimated from the leaves of plants inoculated with third instar nymphs of papaya mealybug and showed a maximum level of crinkling in plants. GA content was higher in infested plants than uninfested plants. The estimated GA content in infested leaves of papaya was 5.0µg/g whereas GA content of 1.0 µg/g was recorded from uninfested leaves of papaya (Table 3).

Table 3. Effect of P. *marginatus* on the gibberellic acid content in papaya seedling

Stage of mealybugs released	Gibberellic acid content in leaves (µg/g)	
	Infested	Uninfested
III instar	5.0	1.0

Mean of 3 replications

The third instar mealybug infested papaya plants showed a fivefold increase in the GA content. There was a direct relationship between the level of damage and GA content. This was in accordance with the result obtained by Saikia *et al*. (2011) that GA_3 content increased after infestation by *Helopeltis* in tea plants. Red spider mite infestation also induced higher level of gibberellic acid in plum trees. Yokomi *et al*. (1995) found that application of chlormequat chloride, a gibberellic acid biosynthesis inhibitor, induced leaf silvering symptoms similar to those induced by the silver leaf whitefly in squash plants.

The gibberellic acids (GAs), a group of diterpenoid compounds with phytohormone activity, affect various stages of plant development, including seed germination, stem elongation, root growth, flowering and pollen tube elongation (Davies, 2004; Swain and Singh, 2005). The above reports also suggest that sucking insects can alter the quantity of GA in infested plants thereby affecting plant growth and development. An excess of GA in infested plants can be considered as a consequence of altered plant metabolism which in turn cause more damage like leaf crinkling and in later stages cause more regrowth.

In the present study it was inferred that third instar papaya mealy bug infestation on papaya significantly reduced the total protein and indole acetic acid content in the plants whereas gibberellic acid content increased significantly. It would help us to know the hypersensitivity of plant species to the infestation of papaya mealy bug, the pest load that a host plant could bear and to evolve appropriate management practices to reduce such reactions.

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(Received 05-02-2015; accepted 19-03-2015)

ENTOMON 39(3): 129-134 (2014)
Article No. ent. 39302

Description of a new species of *Eofoersteria* Mathot (Hymenoptera: Mymaridae) from India, with a key to world species

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ABSTRACT: A new species of the genus *Eofoersteria* Mathot (Hymenoptera: Mymaridae), *Eofoersteria manipurensis* sp. nov., is described from India, a key to the females of all known species and their distribution is provided.

KEYWORDS: Mymaridae, *Eofoersteria*, new species, key, India.

INTRODUCTION

The genus *Eofoersteria* Mathot (1966) (Hymenoptera: Mymaridae) belongs to the *Camptoptera* group of genera. *Eofoersteria* is very minute in size and is similar to *Camptoptera* Foerster with reduced or apparently reduced numbers of antennal and tarsal segments. Currently the genus is represented worldwide by three species (Noyes, 2014), of which only one species, *E. secunda* Viggiani (1978) is known from Tamil Nadu, India. We describe herein a new species from India after three decades, based on a specimen collected in Manipur and provide a key to the world species of *Eofoersteria* along with their distribution.

MATERIALS AND METHODS

Absolute measurements in millimeters are given for body length only. All other measurements are relative, taken with the help of an ocular micrometer having a linear scale of 100 divisions, placed in the eye piece of a compound microscope. All the measurements were made on the same magnification for all the parts. Abbreviations used are: F1 - 6 = Funicle segments 1 - 6; TI - TVI = Tarsal segments I - VI; MHNG – Muséum d'Histoire Naturelle, Geneva, Switzerland; ZDAMU - Insect Collections, Department of Zoology, Aligarh Muslim University, Aligarh, India.

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RESULTS AND DISCUSSION

Genus *Eofoersteria* Mathot

(Figures 1 - 6)

Eofoersteria Mathot, 1966:231 (Type species *Eofoersteria camptopteroides* Mathot, by original designation). Huber & Lin, 1999: 37 - 38.

Diagnosis

Female. Body length, 0.25 - 0.32 mm. Head with median vertical groove extending from foramen to vertex on occiput and transverse groove extending to lateral margin of head below eye. Mandibles unidentate, sharply pointed. Antenna 9-segmented (formula 1161), without ring-like segment (Fig. 1); clava entire. Mesosoma (Fig. 5) with prosternum pointed anteriorly; scutellum without transverse row of fovea; propodeum mostly about half the length of scutellum, carinae present or absent. Fore wing narrow but distinctly curved; proximal macrochaeta absent, distal macrochaeta short. Tarsi 4-segmented (TIV & TV fused). Metasoma (Fig. 5) with ovipositor not exserted to slightly exserted, mostly originates from middle of gaster.

Colour. Body colour dark brown except antennal segments and legs light brown. Fore wing slightly infuscate at base, rest hyaline. Hind wing subhyaline.

Male. Unknown.

Hosts. Unknown.

Distribution. Australia, India, Florida, Trinidad & Tobago, Zaire (Noyes, 2014).

Species. World, 4 species. India, two species (including one new species described herein).

Comments. *Eofoersteria* Mathot can be distinguished from *Camptoptera* Foerster by the funicle 6-segmented, without a ring-like segment and 4-segmented tarsi, with the apical two segments almost fused (in *Camptoptera,* funicle is 7-segmented, with F2 usually ring-like and 5-segmented tarsi, with apical two segments clearly separated forming tarsal segments 4 and 5).

Key to world species of Genus *Eofoersteria,* Females

1. Fore wing with only one median row of discal setae; propodeum with carinae and sculpture(Viggiani, 1978; Fig. 3)………….…....………….......…………………....….………… 2

-. Fore wing with more than two or more rows of discal setae; propodeum smooth (Fig. 9 in Mathot, 1966)…...……………………..……………….............……………..……………3

2. F1 1.3x F2; clava shorter than three preceding funicle segments combined (Fig. 1 in Viggiani, 1978) [India] .....…….…………………………...……............……*secunda* Viggiani

-. F1 0.8x F2; clava longer than three preceding funicle segments combined (Fig. 1) [India] …………...………...……...….......……….………………...............………… *manipurensis* sp. nov.

3. F1 0.7x F2; F2 and F3 subequal and slightly longer than F4 (Fig. 9 in Mathot, 1966) [Democratic Republic of Congo (Zaire)] ………...………………... *camptopteroides* Mathot

-. F1 0.9x F2; F3 longest, F2 subequal to F4 [Australia] ……..........……. *vasta* (Girault, 1920)

1. *Eofoersteria secunda* Viggiani

Eofoersteria secunda Viggiani, 1978: 110–39, Holotype, Female. India. Tamil Nadu (MHNG, not examined). Anis & Rehmat, 2013: 3, checklist. Manickavasagam & Rameshkumar, 2013: 563, checklist.

Diagnosis

Female. Body length 0.32 mm. Body colour brown, with gaster dark brown; antenna and legs light brown. Head with interocullar area with polygonal cells and transverse sculpture; occipital area transversely striated. Antennal scape twice as long as pedicel; F1 longest, F2 subequal to F3, F5 subequal to F6, F4 slightly longer than F5, clava shorter than F4 - F6 combined or three preceding segments. Mesosoma longer than metasoma; pronotum short, hardly visible in dorsal view, mesoscutum with transverse reticulations having polygonal cells; scutellum with more prominent sculpture towards posterior part, metanotum extremely reduced; propodeum with transverse reticulations having 4 carinae medially. Fore wing longer than body, with three rows of discal setae, one row of median discal setae and two submarginal rows. Hind wing with one row of median discal setae. Legs normal, with 4-segmented tarsi. TIV longest. Metasoma with short petiole. Ovipositor slightly longer than metasoma.

Male. Unknown.

Distribution. India: Tamil Nadu.

Comments. The above diagnosis is based on the original description and illustrations given by Viggiani (1978). *E. secunda* differs from to *E. camptopteroides* Mathot in having F1 about 1.3x as long as F2; fore wing with only one row of discal setae and propodeum with carinae. (In *E. camptopteroides* Mathot F1 about 0.7x as long as F2; fore wing with more than two rows of discal setae and propodeum without any carinae.

2. *Eofoersteria manipurensis* sp. nov.

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(Figures 1 - 6)

Description

Female. Body length 0.29 mm. Body colour dark brown except antennal segments and legs light brown. Fore wing with slight infuscation at base, rest hyaline. Hind wing subhyaline. Head (Fig. 4) with occipital area with transverse reticulations having polygonal cells in the middle; posterior ocellus almost touches the trabeculae and ocelli present in an obtuse angle, frontal trabeculae divided into 6 pieces. Mandibles unidentate. Antenna (Fig. 1) with scape about 1.4x as long as pedicel; F1 0.83x of F2 length, F3 - F6 almost equal in length and width, except F6 slightly wider; clava longer than F4 - F6 combined, with 4 longitudinal sensillae covering whole length of clava. Relative measurements: scape length (width) 16 (5); pedicel length (width) 11 (3); F1 length (width) 10 (2); F2 length (width)12 (2); F3 length (width) 8 (3); F4 length (width) 8 (4); F5 length (width) 9 (4); F6 length (width) 9 (5); clava length (width) 29 (10).

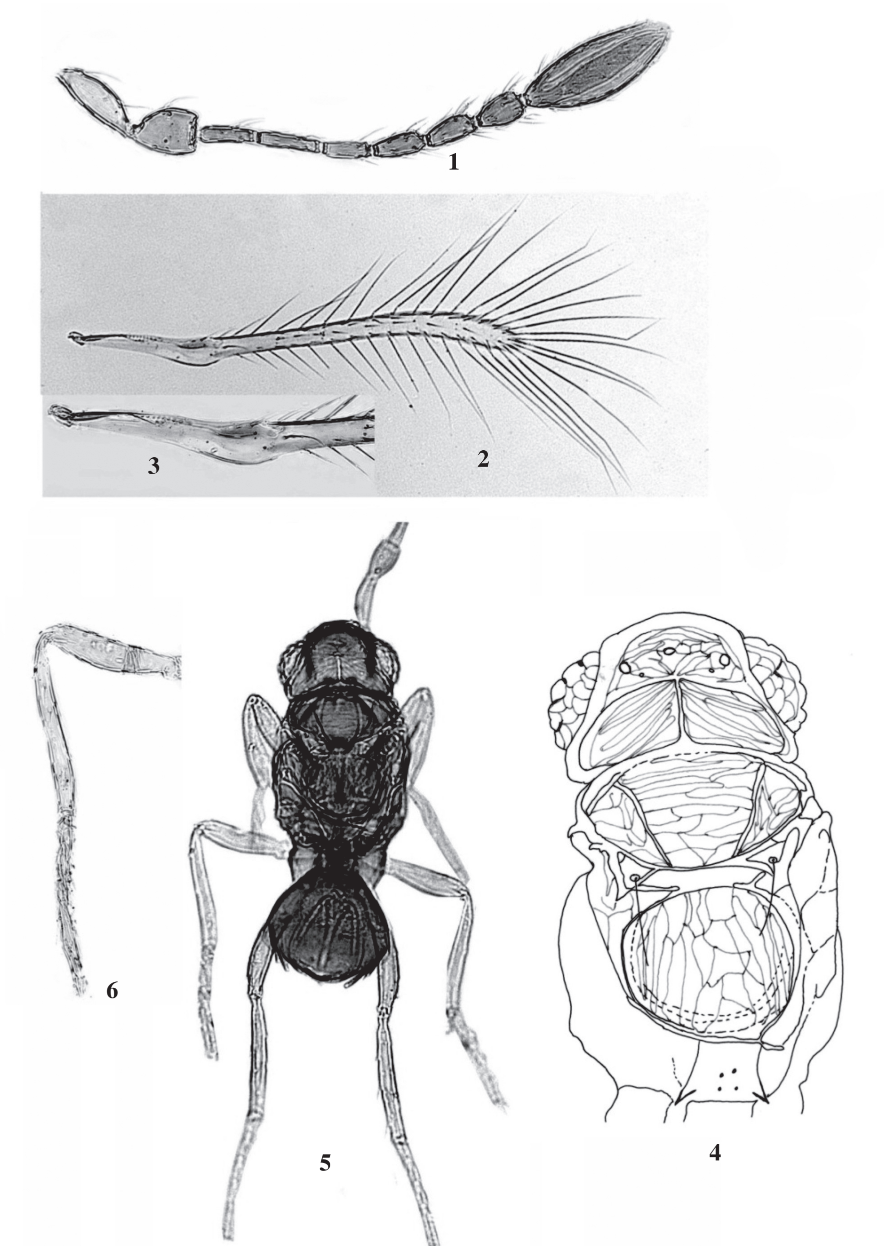
Mesosoma (Fig. 4) longer than metasoma (53:25), with transverse reticulation having longitudinal cells anteriorly and irregular cellular sculpture posteriorly on middle lobe of mesoscutum, lateral lobes with irregular cellulate sculpture; anterior scutellum smooth, posterior scutellum with narrow longitudinal reticulate sculpture laterally and transverse reticulations with large cells medially; each axilla obliquely striate laterally, with a long and strong seta reaching to half of scutellum length; propodeum smooth, with 2 carinae and 4 peg like setae clearly visible between propodeal carinae. Fore wing (Fig. 2) narrow, 18.7x as long as broad; longest marginal fringe, 8.12 x as long as width of fore wing, venation with 2 macrochaeta on marginal vein, 10–11 tubercles and a row of 8 setae on middle of wing disc and a single seta below venation, about 2x as long as other discal setae in middle row of disc. Hind wing 33.7x as long as broad with a row of setae in middle of disc. Legs (Fig. 6) 4-segmented tarsi, TIV & TV fused (genus character), as long as scape length.

Metasoma (Fig. 5) shorter than mesosoma; petiole (Fig. 5) wider than long, sculptured with transverse reticulation; ovipositor not exserted and covers 0.62x of total gaster length. Cercal setae nearly as long as half of gaster length.

Male. Unknown.

Hosts. Unknown.

Material examined. *Holotype* Female (on slide under 4 coverslips), INDIA: MANIPUR: Imphal, 11.11.2011, Coll. S. Begum. (ZDAMU, Registration No. HYM. CH. 712)



Figures 1 - 6. *Eoforesteria manipurensis* sp. nov. Female (Holotype): 1, antenna; 2, fore wing; 3, fore wing with basal part enlarged; 4, head + mesosoma, showing sculpture; 5; body, dorsal; 6, Mid leg.

Distribution. India: Manipur.

Etymology. The species is named after the Indian state Manipur, from where it was collected.

Comments. The species is close to *E. secunda* Viggiani (1978), it differs as follows: F1 shorter than F2 (F2 longest), clava longer than the last three funicular segments combined; propodeum without any reticulations and presence of 4 peg like setae clearly visible in the middle of the carinae; ovipositor 0.62 x of gaster length (see also key to species). (In *E. secunda,* F1 is about 1.3x as long as F2, clava is shorter than three preceding antennal segments together, propodeum with transverse reticulations, but without setae and the ovipositor is slightly longer than the gaster).

ACKNOWLEDGEMENTS

We are grateful to the Chairman, Department of Zoology, Aligarh Muslim University, for providing research facilities. We thank Dr. M. Hayat, Principal Investigator of the ICAR "Network Project on Insect Biosystematics" (NPIB) for his valuable suggestions and support. We gratefully acknowledge the University Grant Commission (UGC), New Delhi, for providing financial assistance.

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(Received 08 January 2015; accepted 13 March 2015)

ENTOMON 39(3): 135-142 (2014)
Article No. ent. 39303

Efficacy of the plant *Centella asiatica*(L.) Urb.(Apiaceae) on *Callosobruchus chinensis* L. (Coleoptera: Chrysomelidae: Bruchinae)

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ABSTRACT: Effect of aqueous, ethanol and acetone extracts of leaves of *Centella asiatica was* tested against the beetle *Callosobruchus chinensis*. The efficacy of the extracts on the test insect was dose-dependent. The results showed that higher doses of the extracts were significantly more toxic to *C. chinensis* compared to lower doses. LD50 value was assessed using probit analysis. After 48 hours of extract exposure of the plant, total body protein, glycogen and total free amino acids and the enzyme transaminase of the insects analysed indicated that total body protein and glycogen was found increased compared to control, while total free amino acid decreased. The enzyme transaminase was found increased. ©2014 Association for Advancement of Entomology

KEY WORDS: *Callosobruchus chinensis,* protein, glycogen, *Centella asiatica*, transaminase, insecticidal activity.

INTRODUCTION

The pulse beetle, *Callosobruchus chinensis* L. (Chrysomelidae: Bruchinae) is a widespread and destructive major insect pest of stored legumes (Park et al., 2003). This insect is considered as a notorious pest of green gram, chickpea, black gram, peas, cowpea, lentil and pigeon pea. Female beetle lays eggs on the seeds. Hatching larvae bore inside and spend their life within the seed. Thus the pulses become completely hollow and unsuitable for human consumption.

Various methods have long been used for controlling insects including the use of chemicals. But pesticides cause toxicity to humans and warm-blooded animals (Salma Mazid and Jogen Ch. Kalita, 2011). There comes the importance of biological pesticides as they do not lead to

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resistance because of their degradable nature. Biopesticides are a group of naturally occurring, often slow-acting protecting agents that are usually safer to humans with minimal residual effects to the environment than conventional pesticides. Biopesticides can be biochemical or microbial.This study is an attempt to analyze the effective repellent and toxicant properties of the plant *C. asiatica* against the stored product pest *C. chinensis*. The test plant being medicinal, would yield environmentally sound chemicals having no harmful effects on the non target organisms. The biological activity of the plant extract might be due to the various compound, including phenolics, terpenoids, and alkaloids, exist in plants (Kashmira et al., 2010). Keeping this in view, the present study was carried out to test the efficacy of the leaf extracts of the plant *C. asiatica*. It is a herbaceous annual plant, growing in tropical swampy areas and has mild anti bacterial, anti viral, anti inflammatory and anti ulcerogenic properties (Newall et al., 1996)

MATERIALS AND METHODS

Culturing of test insects:

Experiments were conducted in the Entomology Research Laboratory, Department of Zoology, University College, Thiruvananthapuram. *C. chinensis* L. adults were obtained from naturally infested green gram seeds from local markets. The adult male and female beetles were reared on clean and un-infested green gram (*Vigna radiata* L). The seeds were made pesticide free by washing with clean water. Three jars each of 300 g were used. Each jar was filled with 200g chickpea grains and about 100 beetles were added to each jar. The jars were then covered.

Preparation of aqueous extract of plants:

The plant leaves were collected and washed well with distilled water. The leaves were ground without adding water. 25 g of the ground mass was then transferred into a beaker containing 100 ml of distilled water. Then it was mixed well and kept for three days. After three days the mixture was filtered. Then this mixture was kept in a water bath at 60-70^{0}C. After drying, this residue is dissolved in water and made up to different concentrations (Talukdar and Howse, 1993).

Preparation of acetone and ethyl alcohol extracts of plants:

For the extraction, soxhlet apparatus was used. 25g powder of plant leaves were extracted with 250ml ethyl alcohol and acetone. The extraction of each plant sample was done in about 12 hrs. After soxhlet extraction; the material was run on rotary evaporator. The extracts were concentrated on rotary evaporator by removing the excess solvent under vacuum. After evaporation of solvent with rotary evaporator the remaining extracted material was kept in a water bath for removing remaining solvent from the extracts. The extracts were stored at 4°C prior to application.

Treatments:

The extracts were applied at different doses (0.2%,0.4%,0.6% and 0.8%) on Whatmann No. 1 filter paper and air-dried for an hour. The controls were treated with acetone, ethyl alcohol or distilled water respectively for acetone, ethanol and aqueous treatments respectively. The treated and control filter paper discs were placed singly at the bottom of plastic jars and 200g of green gram seeds were placed on the papers. Hundred test insects *were* released in each plastic container. There were three replicates for each treatment and control. Observations were recorded on the seventh day of treatment.

Biochemical and enzyme assay:

Bioassay of protein (Lowry et al., 1951), total free amino acids (Spies,1957), glycogen (Dubois et al., 1956) and enzymes transaminases (AsAT [E.C.2.6.1.1.] and AIAT [E.C.2.6.1.2]) (Reitman and Frankel, 1957) were assayed on both control and insects exposed to sub lethal doses of *C. asiatica* .

Electrophoresis of whole body protein of adult beetles subjected to sub lethal dose was conducted according to the method devised by Laemmeli (1970). Electrophorogram obtained was subjected to GEL-DOC analysis using Lab Image Platform software.

Statistical analysis of data:

The data obtained are recorded as mean ± standard deviation. For testing the significance of the data obtained, statistical analysis were carried out using ANOVA (pd"0.05) using SPSS software (Daniel, 2006). LD 50 was calculated using probit analysis (Muhammad Akram Randhawa, 1944).

And standard error of LD 50 was calculated using the formula,

$$\text{Approx. S.E of LD 50} = \frac{\text{Log (LD84-Log LD16)}}{\sqrt{2N}}$$

Probit values were plotted against log doses and the dose corresponding to probit 5 that is 50% was found out.

RESULTS AND DISCUSSION

Effect of plant extracts on mortality of insects:

The total number of adult insects surviving after the treatment was recorded for seven days consecutively. Acetone and Ethanol extracts of the plant showed significant mortality compared to the aqueous extract. No mortality was seen in the case of control. Table 1,2 and

3 shows effect of aqueous, ethanol and acetone extract of plant leaves on LD 50 against the insects of *C. chinensis* respectively.

In the case of aqueous extract maximum mortality was seen in higher doses compared to lower ones (Table 1). In the probit analysis Log LD 50 was 2.8 and LD 50 was 56 mg in the case of aqueous extract.

Table 1. Effect of aqueous leaf extract on LD 50 against adult insects of *C. chinensis*

Group	Dose (wt of plant/wt of feed) mg kg^{-1}	Log dose	% dead	% corrected	probit
1	200	2.3	26	26	4.36
2	400	2.6	45	45	4.87
3	600	2.7	52	52	5.05
4	800	2.9	60	60	5.25

In the case of ethanol extract it was clear that the mortality percentage was more in higher doses compared to aqueous extract (Table 2) and Here Log LD 50 was 2.75 and LD 50 was found as 50.5 mg.

Table 2. Effect of ethanol leaf extract on LD 50 against adult insects of *C. chinensis*

Group	Dose (wt of plant/wt of feed) mg kg^{-1}	Log dose	% dead	% corrected	probit
1	200	2.3	20	20	4.16
2	400	2.6	40	40	4.75
3	600	2.7	50	50	5.00
4	800	2.9	60	60	5.25

In the effect of acetone extract on treated insects the percentage of mortality was higher compared to aqueous and ethanol extracts (Table 3). In the probit analysis Log LD 50 was 2.6 and LD 50 was 48mg. Effectiveness of the extracts was in the order acetone > ethanol > aqueous.

Table3. Effect of acetone leaf extract on LD 50 against adult insects of *Callosobruchus chinensis*

Group	Dose (wt of plant/wt of feed) mg kg^{-1}	Log dose	% dead	% corrected	probit
1	200	2.3	28	28	4.42
2	400	2.6	48	48	4.95
3	600	2.7	56	56	5.15
4	800	2.9	64	64	5.36

Lethal and sub lethal doses of different extracts of the plant *C. asiatica* on mortality of *C. chinensis* were respectively 56mg and 54mg in the case of aqueous extract, 50.5mg and 48.5 mg in ethanol extract and 48 mg and 46 mg in acetone extract (Table 4). 50 percentage of mortality was obtained in lethal doses.

Table4. Effect of sub lethal and lethal dose of aqueous, ethanol and acetone extract of *Centella*

Aqueous extract Dose (mg)	Mortality rate (%)	Ethanol extract Dose (mg)	Ethanol extract rate (%)	Acetone extract Dose (mg)	Mortality rate (%)
(sub lethal) 54	34 ±0.02	(sub lethal) 48.5	38±0.00	(sub lethal) 46	42±0.00
(lethal) 56	50±0.01	(lethal) 50.5	50±0.01	(lethal) 48	50±0.01

Values are mean ±SE; all values are significant at $p \leq 0.05$ level of significance

Effect of plant extracts on bio molecules of insects:

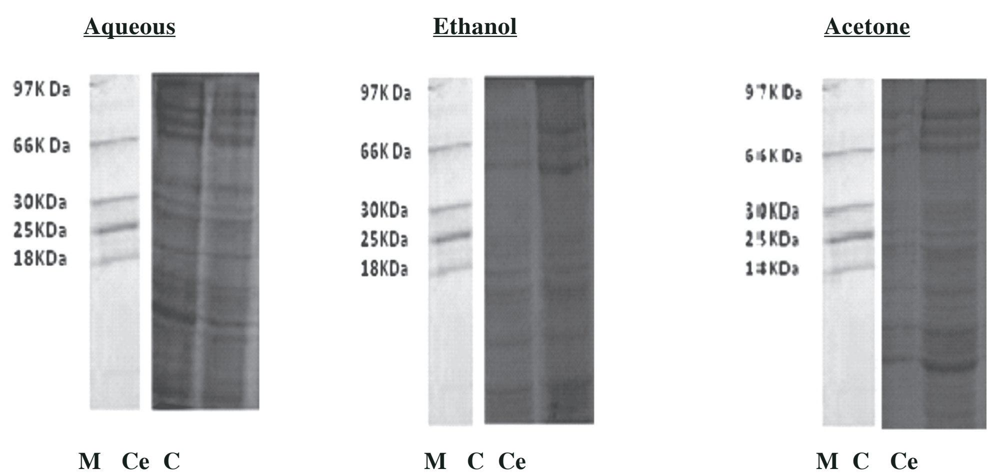
After exposure of sub lethal doses of different extract of the plant extract protein, glycogen and the enzyme transaminase was found to be increased while total free amino acids was found to be decreased when compared to the control insects (Table 5). Electropherogram of aqueous, ethanol and acetone treated insects showed the appearance of new and thick bands compared to control (Fig.1).

The use of plant extracts to control stored products insects is an ancient practice and known to possess bioactive compounds that are either toxic to insects, *Sitophilus zeamais* and *Tribolium castaneum* at various stages of life or elicit anti-feedant properties (Huang et al.,

Table5. Effect of sub lethal dose of different extracts of *C. asiatica* on glycogen, protein, amino acid, GPT and GOT contents in *C. chinensis*

Bio chemical Parameters	Aqueous extract		Ethanol extract		Acetone extract	
	Control insect	Treated insect	Control insect	Treated insect	Control insect	Treated insect
Glycogen ($\mu g\ mg^{-1}$)	0.593±0.002	1.34±0.003	1.34±0.003	1.45±0.002	0.592±0.002	1.73±.006
Protein ($\mu g\ mg^{-1}$)	9.27±0.001	9.40±0.006	9.40±0.006	9.52±0.003	9.28±0.006	10.16±.005
Total free aminoacids ($\mu g\ mg^{-1}$)	9.03±0.004	8.44±0.004	8.44±0.004	8.19±0.001	9.06±0.001	7.84±.002
GPT	5.12±0.021	6.50±0.004	6.50±0.004	7.23±0.024	5.21±0.012	7.60±.004
GOT	3.86±0.011	5.23±0.009	5.23±0.009	5.43±0.031	3.89±0.008	5.69±.003

Values are mean ±SE; all values are significant at $p \leq 0.05$ level of significance



M- Marker **Ce - *Centella asiatica*** **C-Control**

Fig. 1. SDS-PAGE protein patterns of total body of *C. chinensis* treated with aqueous, ethanol and acetone extract of *C. asiatica*

2000). Zia et al. (2011) studied the effect of aqueous extracts from ten plant species on mortality of chickpea beetle, *C. chinensis*. The plant *C. asiatica* contains primary metabolites like saponins also called triterpenoids which are known to be insect deterrent and toxic to insects. Toxic and deterrent modes of action have been suggested as responsible for the activity of several triterpenoid (Ortego et al., 1999). Results obtained in the present investigation clearly demonstrated that both solvent and aqueous extracts of *C. asiatica w*ere toxic to *C. chinensis.* Maximum mortality was obtained in acetone extract of the plant.

Elevated levels of protein indicate the de novo synthesis of novel proteins to overcome stress. Appearance of new protein bands and excess production of proteins as evidenced by SDS PAGE electrophorogram indicated that the insect responded effectively to stress conditions like inability in breathing or feeding deterrence that aroused due to plant extract treatment. Total free amino acids level in the control insect was higher compared to treated insect. The low level of total free amino acids in the test insect indicated a disturbance in the metabolism of insect. This decrease can be because of increased neuromuscular activity of treated larvae which resulted in higher demands for energy. Because of it, high amount of free amino acids entered into the Tricarboxylic acid cycle and oxidized, resulting free amino acid reduction (Chen, 1966). Decrease in free amino acid content indicated the possibility of active catabolism of amino acids through transamination and subsequent entry of corresponding ketoacids into Kreb's cycle to meet the emergent energy requirement as well as their utilization in the production of some new proteins synthesized to cope up with the stress. Hyperglycemia may be due to the inhibition of glycolytic enzymes on exposure to different physical, chemical and biological stress occurred due to treatment. It is also reported that crude plant extracts increase the whole body glycogen content in *S. litura* (Sahayaraj and Agnul, 2004). The increased GOT and GPT activity suggests the mobilization of amino acids during the insecticidal stress exerted by certain components present in the extract to meet the energy demands. Elevation or reduction in enzyme level is associated with physiological imbalance in insects (Saleem and Shakoori, 1985). Changes in the levels of certain enzyme during the course of insecticidal effect of malathion and carbaryl is also reported (Chanda, 1991) .

ACKNOWLEDGEMENT

We express our heartful thanks to University College, Department of Zoology Thiruvananthapuram for providing necessary facilities for the study.

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(Received 04-02-2015: accepted 29-04-2015)

ENTOMON 39(3): 143-150 (2014)
Article No. ent. 39305

Infestation of *Quadrastichus erythrinae* Kim (Eulopidae: Hymenoptera) an invasive pest on *Erythrina* spp, a popular standard for black pepper (*Piper nigrum*) in Idukki district in Kerala, India

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ABSTRACT: Black pepper is an important spice crop grown in Kerala state. Idukki and Wayanad districts contribute major share in production of this spice crop. More than 80 percent of pepper vines grown in this area are trailed on *Erythrina* spp. A survey has been conducted to ascertain the severity of gall wasp infestation in *Erythrina* spp. The total number of locations and black pepper gardens covered during the survey include 6 blocks and 37 panchyats in Idukki district of Kerala state. *E. variegata* (dense thorn) could be rated as moderately resistant (Scale 2- 25% infestation and less) and *E. variegata* (white thorn) as less susceptible (Scale 3 – 25 to 50 % infestation). The reaction of different types/species of *Erythrina* to EWG has revealed that the infestation was higher in black thorn type of *E. variegata* where in the infestation was 75 to 100 percent. Chakkupallam, Erattayar, Kanchiyar of Kattappana panchayats recorded maximum infestation since the pepper growers used black thorned type of *E. variegata* as standards.

KEY WORDS: Black pepper, *Erythria* spp, *Quadrastichus erythrinae*, Erythrina Gall Wasp, Idukki

INTRODUCTION

Black pepper is an important spice crop grown in Kerala state. Idukki and Wayanad districts contribute major share in production of this spice crop. More than 80 percent of pepper vines grown in this area are trailed on *Erythrina* spp. The tree is commonly known as coral tree, tiger's – claw, Japanese coral tree is noted for its seasonal showy red flowers. It is generally

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propagated through cuttings. The tree is commonly preferred as standards for trailing pepper on account of its faster growth, supplementation of nutrients, less competition and presence of thorns which helps in easy trailing by pepper plants. *Erythrina* spp were found affected by various pests and diseases. The most devastating one recorded was Erythrina Gall Wasp (EGW) *Quadrastichus erythrinae* Kim. (Eulopidae: Hymenoptera). Severe incidence of *Quadrastichus erythrinae* has been reported from plains of Thiruvananthapuram on *E. stricta.* (Faizal *et al*. 2006) Outbreak of this pest on *Erythrina* spp was recorded all over Idukki district and the problem has actually felt by farmers of Chellarkovil, Kungiripetty and Myladumpara villages of Chakkupallam panchyat, parts of Udumbanchola and Kattapana block panchyat. (Rajkumar *et al*. 2007). He also reported that black thorned type of *Erythrina variegate* was most susceptible compared to other species. Jacob and Devasahayam (2010) reported incidence of EGW in major black pepper areas of Kerala and Karnataka. The survey indicated that EGW was present in all the districts and taluks surveyed in Kerala and Karnataka.

The incidence and severity of damage caused by EGW significantly varied on various *Erythrina* spp and also on various types of *E. variegata.* Narayana and Dhanya (2011) reported that there was a reduction in the incidence of the pest since the farmers used alternative standards for trailing pepper. An extensive survey in all panchyats of Idukki district for this invasive pest was lacking and hence the survey has been undertaken. The results revealed that the EGW was present in all six blocks and thirty seven panchyats of Idukki district in Kerala. The percentage of trees infested by EGW was higher in Kattapana block (25.4 %) followed by Nedumkandam (23 %) which were in turn on par with Idukki (21.9), Thodupuzha (15.0 %) and Adimali blocks (14.9 %). The thorn less or less thorn type of. *E. subumbrans* was found to be free of infestation by EGW.

MATERIALS AND METHODS

A survey has been conducted during 2010-11 to ascertain the severity of gall wasp infestation in *Erythrina* spp. The total number of locations and black pepper gardens covered during the survey include 6 blocks and 37 panchyats in Idukki district of Kerala state. The black pepper gardens in various panchyats were selected from prominent growers of black pepper who use *Erythrina* spp. as standards for trailing black pepper. From each garden, 15 trees were selected at random and the number of trees with symptoms of damage was recorded and the total number of twigs available and number infested was also recorded to calculate the percentage of trees/twigs infested in each garden. The percentage of trees/ twigs infested was also calculated for each panchyat and each block of Idukki district.

A scale has been developed to measure the infestation of gallwasp. Three species of *Erythrina* namely *E. variegata*, *E. subumbrans* (Thorn less or less thorned) and *E. stricta* were generally used as standards and further three distinct types of *E. variegata* namely, black-thorn type, white-thorn type and white dense-thorn type were also found to occur in different panchayats of Idukki District Kerala India. The reaction of different species/type of *Erythrina* was also studied by developing a scale based on the percent infestation of the twigs by EGW. The rating is as follows.

Scale	Cataegory	Severity of twig infestation
1	Resistant	No infestation
2	Moderately Resistant	25% infestation and less
3	Less Susceptible	25-50 % infestation
4	Susceptible	50 - 75 % infestation
5	Highly Susceptible	75-100 % infestation

RESULTS AND DISCUSSION

The results revealed that the EGW, *Quadrastichus erythrinae* Kim. was present in all six blocks and thirty seven panchyats of Idukki district in Kerala. The percentage of trees infested by EGW was higher in Kattapana block (25.4 %) followed by Nedumkandam (23 %) which were in turn on par with Idukki (21.9), Thodupuzha (15.0 %) and Adimali blocks (14.9 %). The

Table.1 Areas surveyed for the incidence of EGW, *Quadrastichus erythrinae* on *Erythrina* spp.

Block	Panchyat	No. of locations	No. of gardens
Idukki	Arakkulam	5	12
	Kanjikuzhi	4	10
	Kamakshi	7	12
	Mariapuram	5	10
	Vathikudy	4	11
	Vazhatoppu	5	12
Kattapana	Aiyappancovil	5	10
	Chakkupallam	8	15
	Erattyar	8	15
	Kanchiyar	5	15
	Kattappana	8	12
	Upputhara	4	12
	Vandanmadu	2	12

Nedumkandam	Karunapuram	5	10
	Nedumkandam	8	12
	Pampadumpara	8	3
	Rajakkad	5	1
	Rajakumari	5	1
	Senapathy	5	1
	Udumbanchola	8	1
Thodupuzha	Edavetty	5	2
	Karikunnam	5	1
	Kumaramangalam	5	1
	Manakkad	5	1
	Muttom	5	1
	Purapuzha	4	1
Adimali	Adimali	5	2
	Bisonvalley	5	2
	Konnathady	4	2
	Pallivasal	4	2
	Vellathuval	5	2
Azutha	Elappara	5	2
	Kokkayar	5	2
	Kumily	5	2
	Peerumedu	5	2
	Peruvanthanam	5	2
	Vandiperiyar	5	2

percentage of twigs infested by EGW was higher in Kattapana block (15.5 %) followed by Nedumkandam (15.0 %) which were on par with Idukki (14.0 %) and Thodupuzha (11.1 %) (Table 2).

Table. 2 Incidence of EGW, *Quadrastichus erythrinae* on *Erythrina* spp in major black pepper growing blocks of Idukki District.

Blocks	Mean per cent infestation	
	Tree	Twig
Idukki	21.9^{ab}	14.0^{ab}
Kattapana	25.4^{b}	15.5^{b}
Nedumkandam	23.8^{b}	15.0^{b}
Thodupuzha	15.0^{ab}	11.1^{ab}
Adimali	14.9^{ab}	10.0^{a}
Azutha	13.7^{a}	9.7^{a}

Means followed by the same letter are not significantly different in Duncans test P = 0.01

Among the panchyats, the percentage of trees infested by EGW was significantly higher in Chakkupallam (37.2 %) followed by Udumbanchola (35.6), Erattayar (35.3 %) and Karunapuram panchyats (32.3 %). The trees infested by EGW was significantly lower in Rajakumari Panchyat (10.2 %). The percentage of twigs infested by EGW was significantly higher in Erattayar (27.8 %) followed by Udumbanchola (22.6 %), and Chakkupallam (22.2 %) which were on par in all other panchyats except Bison Valley (6.8 %), Peerumedu (7.8 %), Peruvanthanam (8.0 %), Purapuzha (8.3 %), Manakkad (9.2 %) and Kumuly 9.3% (Table 4). Rajkumar *et al.*(2007) reported an outbreak of pest on *Erythrina* spp all over Idukki district and severe in Chellarkovil, Kungiripetty and Myladumpara villages of Chakkupallam panchyat, parts of Udumbanchola and Kattapana block panchyat. Jacob and Devasahayam (2010) reported incidence of EGW in major black pepper areas of Kerala and Karnataka. The survey indicated that EGW was present in all the districts and taluks surveyed in Kerala and Karnataka.

The reaction of different types/species of *Erythrina* to EWG has revealed that the infestation was higher in black thorn type of *E. variegata* where in the infestation was 75 to 100 percent. This was in accordance with the findings of Rajkumar *et al.* (2007). The pest infestation was less than 25 per cent in dense thorn type of *E. variegata.* The infestation was 25 to 50 percent in the white thorn type of *E. variegata.* The thorn less or less thorn type of. *E. subumbrans* was found to be free of infestation by EGW. Jacob and Devasahayam (2010) reported the incidence and severity of damage caused by EGW significantly varied on various *Erythrina* spp and also on various types of *E variegata.*

The rating of resistance / susceptibility of various species/ types of *Erythrina* to EGW based on the percentage of damaged twigs indicating that *E. subumbrans* thorn less type could be

Table. 3 Reaction of *Erythrina* spp to *Quadrastichus erythrinae*

Scale	Cataegory	Severity	Species/Type
1	Resistant	No infestation	*E. subumbrans* (Thorn less/ Less thorn)
2	Moderately Resistant	25% infestation and less	*E. variegata* (Dense thorn)
3	Less susceptible	25-50 % infestation	*E. variegata* (White thorn)
4	Susceptible	50 - 75 % infestation	Nil
5	Highly Susceptible	75-100 % infestation	*E. variegata* (Black thorn)

Table. 4 Incidence of EGW, *Quadrastichus erythrinae* on *Erythrina* spp in major black pepper growing panchyats

Block	Panchyat	Per cent mean Damage	
		Tree	Twig
Idukki	Arakkulam	23.5 ab	13.2 ab
	Kanjikuzhi	28.2 ab	18.5 ab
	Kamakshi	12.5 a	10.6 ab
	Mariapuram	23.5 ab	13.5 ab
	Vathikudy	25.6 ab	15.6 ab
	Vazhatoppu	18.2 a	12.2 ab
Kattapana	Aiyappancovil	27.2 ab	12.5 ab
	Chakkupallam	37.2 b	22.2 b
	Erattyar	35.3 b	27.8 b
	Kanchiyar	22.3 ab	13.6 ab
	Kattappana	25.5 ab	10.2 ab
	Upputhara	15.3 a	12.3 ab
	Vandanmadu	15.3 a	10.2 ab

Nedumkandam	Karunapuram	32.3^{b}	13.6^{ab}
	Nedumkandam	28.9^{ab}	18.5^{ab}
	Pampadumpara	25.6^{ab}	12.6^{ab}
	Rajakkad	13.6^{a}	10.3^{ab}
	Rajakumari	10.2^{a}	12.1^{ab}
	Senapathy	20.1^{ab}	15.2^{ab}
	Udumbanchola	35.6^{b}	22.6^{b}
Thodupuzha	Edavetty	23.2^{ab}	11.2^{ab}
	Karikunnam	13.3^{a}	12.1^{ab}
	Kumaramangalam	13.6^{a}	10.3^{ab}
	Manakkad	12.6^{a}	9.2^{a}
	Muttom	12.3^{a}	15.6^{ab}
	Purapuzha	15.2^{a}	8.3^{a}
Adimali	Adimali	23.5^{ab}	10.6^{ab}
	Bisonvalley	14.5^{a}	6.8^{a}
	Konnathady	13.5^{a}	10.6^{ab}
	Pallivasal	15.6^{a}	11.6^{ab}
	Vellathuval	22.4^{ab}	10.2^{ab}
Azutha	Elappara	12.3^{a}	10.5^{ab}
	Kokkayar	15.6^{a}	12.5^{ab}
	Kumily	13.3^{a}	9.3^{a}
	Peerumedu	15.3^{a}	7.8^{a}
	Peruvanthanam	12.6^{a}	8.0^{a}
	Vandiperiyar	13.3^{a}	10.3^{ab}

Means followed by the same letter are not significantly different in Duncans test P = 0.01

classified as resistant (Scale 1- No infestation) and *E. variegata* (black thorn type) highly susceptible (Scale 5- Highly susceptible) 75-100 % infestation. *E. variegata* (dense thorn) could be rated as moderately Resistant (Scale 2- 25% infestation and less) and *E. variegata* (white thorn) as less susceptible (Scale 3 – 25 to 50 % infestation).

Chakkupallam, Eratatyar, Kanchiyar of Kattappana panchayats recorded maximum infestation since the pepper growers used black thorned type of *E. variegata* as standards. Messing *et al* (2008) screened 71 species of *Erythrina* in Hawaii to EGW infestation based on field observations and sleeve- cage experiments, and found that 12 species were free from attack. It has been observed that EWG is an important limiting factor for successful cultivation of black pepper since *Erythrina* sp with thorns were ideal for trailing black pepper. In many gardens the infestation of EGW along with borer pest has resulted in death of *Erythrina* standards. Pepper growers were reluctant in using alternate standards and this has resulted in decline in area of black pepper in many areas of Idukki district.

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(Received 06 December 2014; accepted 30 April 2015)

Entomon 39(3): 151-160 (2014)
Article No. ent. 39306

Insect pests of cabbage and cauliflower and their natural enemies in agro ecosystem of Kerala

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ABSTRACT: Pests and natural enemies occurring on cool season vegetable crops, cabbage and cauliflower, grown in hill tracts and the newly cultivated plains of Kerala were studied. Analysis of fauna showed that these crops were attacked by eleven pests. The occurrence of pierid butterfly, *Appias lyncida* Cramer on cruciferous crops is reported for the first time. Incidence of flea beetle, *Phyllotreta chotanica* Duv. noted during the seedling stage is the first report from these crops. The key pest affecting the crop grown in plains was the cut worm, *Spodptera litura* (F.) and in the hilly tracts it was Diamond back moth (DBM) *Plutella xylostella* (L.). Damage in terms of yield loss by *S. litura* was 30 per cent in cabbage and cauliflower and that by *P. xylostella* was 38 per cent to heads and 26 per cent to curds. Two coccinellid predators, *Chilomenes sexmaculata* (F.) and *Coccinella transversalis* (F.) and the syrphid, *Ischiodon scutellaris* (F.) were identified from the colonies of aphid, *Lipaphis erysimi* (Kaltenbach). One parasitoid, *Protapanteles* sp. was identified from larvae of *Plusia signata* (F.). ©2014 Association for Advancement of Entomology

Key words: cabbage and cauliflower pests, incidence, extent of damage, Kerala, *Spodoptera litura*, *Plutella xylostella*, *Appias lyncida*, *Phyllotreta chotanica*

INTRODUCTION

Cabbage (*Brassica oleracea* L. var. *capitata*) and cauliflower (*Brassica oleracea* L. var. *botrytis*) are the two important cool season vegetables widely grown in all parts of India. In Kerala, cultivation of cabbage and cauliflower was earlier restricted to the cooler seasons in the hilly tracts. With the introduction of new tropical varieties it has become popular in the plains too. Being succulent, these crops are severely attacked by many pests. Bonnemaison (1965) reviewed the distribution of pests attacking crucifers all over the world and reported nearly 51

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insect species. In India, a total of 37 insect pests have been reported to feed on cabbage (Lal, 1975). The diamondback moth (DBM), *Plutella xylostella* (L.), cut worm, *Spodoptera litura* (F.) cause appreciable loss (Bhalani, 1989). Incidence of these pests varies from season to season (Sachan and Gangwar, 1990) and region to region (Chaudhuri *et al.,* 2001). Pests infesting these crops in agro ecosystems of Kerala is being documented in this paper for the first time.

MATERIALS AND METHODS

Identification of pests and natural enemies

The occurrence of pests and natural enemies in cabbage and cauliflower grown in the plains and hilly tracts was monitored during 2011-13. The pest attack was recorded from four districts viz. Thiruvananthapuram, Kollam and Thrissur, representing the plains and from Idukki district in the hilly tracts. From each district five plots having two cents or more were selected. The period of observation was from November to February that coincided with the peak cultivation of these crops in Kerala. Pests attacking the crops at different growth stages were recorded. The distinct growth stages of cabbage and cauliflower described by Andaloro *et al.* (1983) as given under was utilized for this purpose.

Crop specific growth stages

Cabbage		Cauliflower	
WAP	**Specific stage***	**WAP**	**Specific stage***
1 – 4	Seedling stage	1 – 4	Seedling stage
5- 7	True Leaf stage	5- 7	True Leaf stage
8	Pre cupping stage	8- 10	Curd initiation stage
9 – 10	Cupping stage	11 – 13	Curd development stage
11- 12	Early head formation stage	14 - 15	Curd maturity stage
13-14	Head fill stage	-	-
15	Head maturity stage	-	-

WAP- Weeks after planting

The adult and immature stages of the pests were collected from field and brought to the laboratory for identification. The unknown species were identified with the help of experts in the concerned field.

Natural enemies

The predators were collected from the field and were identified and preserved as dry specimens. Parasitised insects were brought to the laboratory and maintained for adult emergence. The adults preserved in 70 percent alcohol were sent for identification.

Extent of infestation and damage caused by pests

From the selected district five plots, each having 2 cents or more were selected for and Pest Infestation Index (PI) was calculated using the formula

PI = Number of plants infested /total number of plants observed x 100

Extent of damage caused by the major pests during the vegetative stage was worked out by counting the total number of leaves and the number of leaves damaged per plant. During the reproductive phase it was calculated by counting the number of heads or curds damaged out of the thirty observational plants. Mean number of leaves and heads or curds damaged was worked out using standard deviation.

RESULTS

Pests

The pests encountered during different growth stages, plant parts affected, susceptible stage of the crop and other relevant information are detailed in Table 1. Eleven insect pests from the order Lepidoptera, two each from Hemiptera and Orthoptera and one each from Diptera and Coleoptera were observed. An unidentified species of slug was also recorded.

Predators

Predators were observed only in fields where there was aphid infestation. The natural enemies recorded from the colonies of *Lipaphis erysimi* were the coccinellid beetles, *Chilomenes sexmaculata* (F.) and *Coccinella transversallis* (F.) . The syrphid predator collected was identified as *Ischiodon scutellaris* (F.).

Parasitioids

Larvae of *P. signata* were found to be parasitised by a braconid endo-parasitiod, *Protapanteles* sp..

Extent of infestation and damage caused by pests

The extent of infestation caused by different pests in different districts was worked out based on the Pest infestation Index (PI). In Thiruvananthapuram, Kollam and Thrissur districts, the major infestation was that of *S.litura*, the PI were 71.99, 71.33, and 69.99 respectively (Table 2). Infestation of other pests in these districts was very low. The minor pests, mustard aphid, *L. erysimi,* semi loopers, *Plusia.signata* (F.), *Plusia orichalcea* (F.), hairy caterpillars, *Pericallia ricini* (F), *Dasychira mendosa* Hb. *Spilosoma obliqua* (Walker) and leaf miner, *Liriomyza ttrifolii*(Burgen), were also prevalent in all the above districts. Their pest infestation index ranged from 8.66 to 12.66, 6.66 to 11.99, 1.33 to 2, 2.66 to 3.99, 2 to 5.33, 2.66 to 4.66 and 7.33 to

Table 1. Details of the pests infesting cabbage and cauliflower in Kerala

Sl No.	Pests		Family	Destructive stage	Susceptible stage of host plant	Plant parts affected
	Common name	Scientific name				
Order: Lepidoptera						
1	Cut worm	*Spodoptera litura*(Fabricius)	Noctuidae	Caterpillar	All stages except seedling stage	Leaves,Head, Curd
2	Diamondback moth	*Plutella xylostella*(Linnaeus)	Plutellidae	Caterpillar	All stages	Leaves, Head, Curd
3	Semilooper	*Plusia signata* Fab.	Noctuidae	Caterpillar	True leaf stage, Precupping and cupping/curd initiation stages	Leaves
4	Semilooper	*Plusia orichalcea* Fab.	Noctuidae	Caterpillar	True leaf stage, Precupping and cupping/curd initiation stages	Leaves
5	Hairy caterpillar	*Dasychira mendosa* Hb.	Lymantriidae	Caterpillar	True leaf stage	Leaves
6	Hairy caterpillar	*Spilosoma obliqua* Walker	Arctiidae	Caterpillar	True leaf stage	Leaves
7	Hairy caterpillar	*Pericallia ricini* F.	Arctiidae	Caterpillar	True leaf stage	Leaves
8	Beet army worm	*Spodoptera exigua* Hb.	Noctuidae	Caterpillar	True leaf stage	Leaves
9	Pierid butterfly	*Appias lyncida* (Cramer)	Pieridae	Caterpillar	True leaf stage	Leaves
10	Bag worm	Unidentified	Psychidae	Caterpillar	Seedling stage	Leaves
11	Bell moth*	Unidentified	Tortricidae	Caterpillar	Early head formation Stage	Growthprimordia

Order: Hemiptera						
12	Mustard aphid	*Lipaphis erysimi*(Kaltenbach)	Aphididae	Nymph, Adult	True leaf stage, Precupping, Cupping/ curd initiation and Early head formation stage	Leaves, Head
13	Cabbage aphid	*Brevicoryne brassicae* (Linnaeus)	Aphididae	Nymph, Adult	True leaf stage, Precupping, cupping/ curd initiation and Early head formation stage	Leaves, Head
Order: Diptera						
14	Leaf miner	*Liriomyza trifolii* (Burgen)	Agromyzidae	Caterpillar	Seedling, True leafStage	Leaves
Order: Coleoptera						
15	Flea beetle	*Phyllotreta chotanica* Duv.	Alticidae	Adult	Seedling, True leafStage	Leaves,Roots
Order: Orthoptera						
16	Short horned Grasshopper	*Atractomorpha crenulata* (Fabricius)	Pyrgomor-phidae	Nymph, Adult	Seedling, True leafStage	Leaves
17	Long horned grasshopper	Unidentified		Nymph, Adult	Seedling, True leafStage	Leaves
Phylum: Mollusca						
18	Slugs*	Unidentified			Early head formationand head fill stage	Head

*Pest of cabbage only

Table 2. Extent of pest infestation in cabbage and cauliflower grown in different districts of Kerala

Sl. No.	Pests	Mean pest infestation index			
		Plains			Hilly tract
		Thiruvananthapuram	Kollam	Thrissur	Idukki
1	*Spodoptera litura*	71.99	71.33	69.99	—
2	*Plutella xylostella*	—	—	—	73.99
3	*Plusia signata*	6.66	11.99	10.66	—
4	*Plusia orichalcea*	2	1.33	1.33	—
5	*Spilosoma oblique*	3.33	4.66	2.66	1.33
6	*Dasychira mendosa*	2	5.33	3.99	2.66
7	*Pericallia ricini*	3.33	3.99	2.66	3.33
8	*Spodoptera exigua*	1.33	—	1.33	—
9	*Appias lyncida*	1.33	—	—	—
10	*Lipaphis erysimi*	9.99	8.66	12.66	—
11	*Brevicoryne brassicae*	—	—	—	19.33
12	*Phyllotreta chotanica*	14.66	7.33	—	—
13	*Liriomyza trifolii*	7.33	15.33	8.66	11.99
14	Grass hoppers	7.99	8.66	9.99	7.33
15	Bell moth	4.66	7.33	10.66	—
16	Bag worm	2.66	3.99	1.33	—
17	Slug	4.66	—	—	11.33

Mean of five locations per district

15.33 respectively. The incidence of flea beetle, *Phyllotreta chotanica* Duv. was observed only in Thiruvanathapuram (PI 14.66) and Kollam districts (PI 7.33). It was not observed in Thrissur district. The pierid butterfly *A. lyncida* was observed only from one location at Thiruvanathapuram district. Other pests with lesser indices were, *Spodoptera exigua (*Hb.), *Atractomorpha crenulata* (F.) and unidentified species of bell moth, bag worm and long horned grass hopper.

The PI of DBM was high (73.99) in the major cool season vegetable growing areas *viz.* Devikulam, Vattavada and Kanthalloor comprising the hilly tracts of Idukki district. In these

areas, infestation index of *S. litura* was zero. The cabbage aphid, *Brevicoryne brassicae* was observed in the hilly tracts only with a PI of 19.33. Other pests observed with less infestation indices were *D. mendosa, S. obliqua, P. ricini, L. trifolii* and *A. crenulata.*

The PI of DBM was high in the major cool season vegetable growing areas viz. Devikulam, Vattavada and Kanthalloor comprising the hilly tracts of Idukky district. In these areas infestation index of *S.litura* was zero. The cabbage aphid *Brevocoryne brassicae* (L.) was observed in the hilly tracts only with a PI of 19.33.The pests that were found to occur in both the hilly tracts and plains were *L. trifolii, D. mendosa, P. ricini, S. obliqua* and *A.crenulata*.

Extent of damage caused by major pests

Spodoptera litura

In cabbage, (Table. 3) the mean number of leaves damaged per plant ranged from 1.4 ± 1.77 to 2.90 ± 1.79. Percentage of leaves damaged during vegetative phase was estimated as 15.32 only but in the bearing stage it caused 30.00 per cent damage to heads. In cauliflower (Table. 4), the mean number of leaves damaged per plant, ranged from 3.2 ± 1.81to 3.7 ± 1.63 and the per cent damage caused to leaves was 20.80 only but in the bearing stage the damage caused to curds was 30 per cent.

Plutella xylostella

The mean number of leaves damaged per cabbage plant ranged from 2.1 ± 1.52 to 2.9 ± 1.52. Percentage of leaves damaged during vegetative phase was estimated as 17.58. In the bearing stage it caused 38 per cent damage to heads.

In cauliflower (Table. 4), the mean number of leaves damaged ranged from 2.4 ± 1.71 to 3.3 ± 1.88. Percentage of leaves damaged during vegetative phase was estimated as 17.63 and the extent of damage caused to curds was 26 per cent.

DISCUSSION

The occurrence of the pierid butterfly, *Appias lyncida* Cramer on cabbage is reported for the first time**.** Earlier this species was not reported from Brassicaceae plants. The pest was collected from Thiruvananthapuram district, where it was found to defoliate cabbage. Later it was observed to feed on cauliflower too when reared in laboratory. Originally the species was reported to feed on plants belonging to the family Capparidaceae only. The observation on the dominance of *S. litura* in plains during the study period of November to February is in agreement with the findings of Murthy (1994), who reported that November to March was the ideal season of multiplication of *S. litura* in warmer climates where cabbage was grown. It was reported as a pest that caused extensive damage to cabbage and cauliflower in Bihar (Chand and Triparthi, 2008), in Dapoli (Ambekar *et al*., 2009). *S.litura* was reported to be limiting

Table 3. Extent of damage caused by *Spodoptera litura* and *Plutella xylostella* during vegetative and reproductive stages of cabbage in Kerala

Location	Mean number of leaves/plant		Mean number of leaves damaged/plant		Extent of leaves damaged (%)		Number of heads damaged		Extent of heads damaged (%)	
	S. litura	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*
1	11.2	13.3	1.6 ± 1.26	2.1 ± 1.72	14.28	15.78	3 ± 0.48	3 ± 0.48	30	30
2	12.5	13.6	2 ± 1.76	2.7 ± 1.70	16	19.85	2 ± 0.42	5 ± 0.52	20	50
3	13.9	15.1	1.4 ± 1.77	2.9 ± 1.52	10.07	19.20	3 ± 0.48	5 ± 0.52	30	50
4	14.6	14.9	2.9 ± 1.79	2.9 ± 1.44	19.86	19.46	3 ± 0.48	3 ± 0.48	30	30
5	13.4	15.4	2.2 ± 1.47	2.1 ± 1.52	16.41	13.63	4 ± 0.51	3 ± 0.48	40	30
Mean	—	—	—	—	15.324	17.58	—-	—-	30	38

Mean of ten plants

Table 4. Extent of damage caused by *S. litura* and *P. xylostella* during vegetative and reproductive stages of cauliflower in Kerala

Location	Mean number of leaves/plant		Mean number of leaves damaged/plant		Extent of leaves damaged (%)		Number of curds damaged		Extent of curds damaged (%)	
	S. litura	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*	*S. litura*	*P. xylostella*
1	17.3	16.2	3.6 ± 2.06	3.3 ± 1.88	20.80	20.37	3 ± 0.48	3 ± 0.48	30	30
2	15.9	15	3.2 ± 1.81	2.6 ± 1.83	20.12	17.33	2 ± 0.42	2 ± 0.42	20	20
3	16	15.6	3.3 ± 1.88	3.1 ± 2.46	20.62	19.87	3 ± 0.48	4 ± 0.51	30	40
4	16.9	15.8	3.7 ± 1.63	2.4 ± 1.71	21.89	15.18	3 ± 0.48	3 ± 0.48	30	30
5	17	14.9	3.5 ± 2.01	2.3 ± 1.25	20.58	15.43	4 ± 0.51	1 ± 0.31	40	10
Mean	--	---	----	---	20.80	17.63	---	----	30	26

Mean of ten plants

factor for successful cultivation of crucifers (Singh *et al.* 2012; Ratnasri 2012).The finding that *P. xylostella* as the key pest in Vattavada, Kaanthallur and Devikulam of Idukki district which is at a higher altitude of 1980 M above mean sea level, is substantiated with the earlier observations made by Saucke *et al.* (2000). There are numerous reports that reveal *P. xylostella* as the key pest of cabbage and cauliflower in various part of the country (Chaudari *et al*. 2001; Mishra 2009; Kumar et al. 2011). Most of the studies were confined to the cooler seasons of the year, which coincided with the major growing season of cabbage and cauliflower.

Among the minor pests, the infestation index was higher (14.66 per cent) for the flea beetle *Phyllotreta chotanica* Duv., but during the study, was under control with the use of any of the botanical or synthetic insecticides. Its incidence was more in non weeded plots. Though it is reported from India for the first time, earlier reports are there on its incidence in kale, *Brassica oleracea* L. from south east Asia (Kianmeesuk *et al*., 1999; Kianmatee and Ranamukhaarachchi, 2007). The other pests observed during this study with lesser infestations were, the hairy caterpillars, *Pericallia ricini* F., *Spilosoma obliqua* Walker, *Dasychira mendosa* Hb, beet army worm, *Spodptera exigua* Hb., and semi looper caterpillars, *Plusia signata* Fab. and *P. orechalcea* Fab. were earlier reported in India, by various workers, as cited in the check list of vegetable pests of India prepared by Sharma (2011). The aphid species observed in plains differed from that seen in hilly areas. *B.brassicae* was seen only in hilly tracts, whereas *L.erysimi* was the species observed in plains.

During the course of this investigation, predators were observed only from those fields where aphid infestation was there. The present study was in conformity with the Anjumoni *et al.* (2011). They reported that the predominant coccinellid species in *L. erysimi* colonies is *C. septumpuncata*.

The extent of damage caused by *S. litura* to the heads and curds was 30 per cent in both cabbage and cauliflower, but the population levels recorded during the two consecutive seasons indicated the crop preference of *S. litura* to cauliflower. However the damage caused by *P. xylostella* varied in these two crops. It was higher in cabbage (38 per cent) when compared to cauliflower (26 per cent). Such studies on comparative susceptibility of these two crops to two different major pests observed at two different altitudes were not seen carried out earlier.

ACKNOWLEDGEMENTS

The authors are grateful to Dr. Kumar Ghorpade, Emeritus Scientist, UAS Dharwad, Dr. Krushnamegh Kunte, National Centre for Biological Sciences, Bengaluru, Dr. Sunil Joshi, Entomologist, National Bureau of Agriculturally Important Insects, Bengaluru, the late Dr. T.C. Narendran,, Taxonomist, Calicut University and Dr. Prathapan, K.D, Taxonomist, Department of Entomology, College of Agriculture, Vellayani for identification of insect specimens.

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(Received 22 April 2015; approved 16 May 2015)

ENTOMON 39(3): 161-169 (2014)
Article No. ent. 39304

Annotated checklist of Indian Ceutorhynchinae (Coleoptera: Curculionidae)

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ABSTRACT: A checklist to all the known 13 genera and 45 species of the subfamily Ceutorhynchinae from India is given. Taxonomic history with details of synonyms, references and distribution are also included. The zoogeographic distribution has been analyzed and details included. ©2014 Association for Advancement of Entomology

Key words: Ceutorhynchinae, India, checklist, distribution

INTRODUCTION

The subfamily Ceutorhynchinae was erected by Gistel (1856) as new placement from Barididae (Alonzo-Zarazaga and Lyal, 1999). This subfamily contains ca.1316 species, and one of the most speciose in the family Curculionidae (Colonnelli, 2004) and itself one of the largest family of Order Coleoptera (Alonso-Zarazaga and Lyal, 1999). Ceutorhynchines are distributed worldwide except New Zealand, Oceania, Antartic and Subantartic regions and Soth America, south of middle Argentina; no species known from Chile; the greatest numbers of genera and species are known from the Palaeartic, followed by Oriental region (Korotyaev, 2006).

Ceutorhynchinae are easily recognized by their robust complexion and ability to place the rostrum between the coxae in repose. One of the main external structural characters are the dorsally visible apices of the mesepimera (except for the Palaeotropical genus *Cyphosenus* Schultze, 1899 and Oriental *Ceutorhynchoides* Colonnelli, 1979). More than one third of the existing Ceutorhynchinae are monophagous or oligophagous on Brassicaceae species (Korotyaev, 2006). Several *Ceutorhynchus* are among the most important pests on cruciferous crops as their larvae and adults damage plants. Larvae of members of this genus tunnel into stems (*Ceutorhynchus napi* Gyllenhal 1837, *Ceutorhynchus pallidactylus* (Marsham 1802)

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and leaf stalks (*Ceutorhynchus picitarsis* Gyllenhal 1837), feed on seeds within pods of the host plants (*Ceutorhynchus obstrictus* (Marsham 1802)) or cause root-gall formation (*Ceutorhynchus assimilis* (Paykull 1800)). *Ceutorhynchus* adults gnaw leaves, stems, buds and flowers of the cruciferous plants (Korotyaev, 2006). Some Ceutorhynchinae species feed on weeds and therefore are used or considered as biological control agents against weeds (Peschken and Wilkinson, 1981; Burki *et al*., 2001; Colpetzer *et al*., 2004; Korotyaev, 2006; Newman *et al*., 2006; Gerber *et al*., 2007; Visalakshy, 2007). There are some interesting biological and morphological peculiarities that occur in subgroups of Ceutorhynchinae and noteworthy is the ability to leap, expressed to a varying degree in several tribes of Ceutorhynchinae, which is not developed to a comparable extent in other Curculionidae except for Rhamphini (Curculioninae) and has been newly acquired within the Ceutorhynchinae (Korotyaev, 2006). Besides, there are aquatic Ceutorhynchinae with well developed swimming abilities and morphological adaptations compared to Bagoinae, the only other subfamily of the Curculionidae with specialized aquatic forms.

MATERIAL AND METHODS

This list is mainly based on available literature rather than on extensive taxonomic studies. It has been compiled mainly with the aid of Zoological Record (Insecta), the "Catalogue of Ceutorhynchinae of the World, with a key to genera" (Colonnelli, 2004) and original descriptions, wherever available. The classification proposed by Schultze (1902); Wagner (1938a, 1938b) and Colonnelli (2004) was followed.

RESULTS AND DISCUSSION

The results revealed that, the Ceutorhynchinae of India are represented by 45 species distributed in 13 genera under five tribes. A perusal of the checklist indicates that majority of the species are distributed in Northeast and north Indian peninsula. An analysis of described Indian fauna indicates that, from 1837 to 1900 only six species had been described (Table 1); 18 species were described from 1901 to 1950. The majority of species (21) were described during 1951 to 2000, and thereafter, no species were described from India. An analysis made on the contributions of coleopterists (Table 2) indicates that Pajni and Kohli had described 10 species followed by Colonnelli (1979, 1984, 1987, 1992, 1993, 1998); and Marshall with nine species each, Hustache with 6 species, Schultze with three species, Korotyaev with two species, and Dalla Torre, Faust, Gyllenhal, Motschulsky with one species each. In conclusion, the present checklist is a comprehensive list of Ceutorhynchines distributed in India.

TRIBE, GENERA AND SPECIES		DISTRIBUTION
A.	**Phytobinii Gistel, 1848**	
I.	*Rhinoncus* Schoenherr, 1825: c586 Type species: *Curculio pericarpius* Linnaeus, 1758	
1.	*caesareus* Colonnelli, 1979b: 483	North Indian peninsula: India, Nepal, Pakistan

2.	*paganus* Gyllenhalin Schoenherr, 1837: 586 *Rhinoncus luzonicus* Hustache, 1925: 333; Colonnelli, 2004: 32	Southeast Asia, South Africa: Bangladesh, Indonesia, India, Laos, Nepal, Philippines, Thailand, Vietnam, South Africa
B.	**Ceutorhynchini Gistel, 1848**	
II.	*Ceutorhynchoides* Colonnelli, 1979a: 144 Type species: *Ceutorhynchoides badius* Colonnelli, 1979b	
3	*bengalensis* Colonnelli, 1984: 201	Bengal: India
4	*notatus* Colonnelli, 1984: 202	Bengal: India
5	*subcostulatus* (Hustache, 1920) *Ceuthorrhynchus subcostulatus* Hustache, 1920: 332; Colonnelli, 2004: 36	South India: India
6	*topali* Colonnelli, 1984: 202	Assam: India
III.	*Ceutorhynchus* Germar, 1824 *Ceutorhynchus* Germar, 1824: 217 Type species: *Curculio assimilis* Paykull, 1792	
7	*kashmirensis* Korotyaev, 1980: 144	Kashmir: India
IV.	*Dieckmannius* Colonnelli, 1987: 139 Type species: *Ceuthorrhynchus sexnotatus* Schultze, 1899	
8	*ingens* Colonnelli, 1993: 388	Himalayas: Bhutan, India, Nepal
9	*sexnotatus* (Schultze, 1899) *Ceuthorrhynchus sexnotatus* Schultze, 1899: 190; Colonnelli, 2004: 56 *Ceuthorrhynchus camerunensis* Hustache, 1930: 78; Colonnelli, 2004: 56	Himalayas: North India, Nepal
V.	*Indicoplontus* Colonnelli, 1984: 205 Type species: *Ceuthorrhynchus indicus* Hustache, 1930	
10	*indicus* (Hustache, 1930) *Ceuthorrhynchus indicus* Hustache, 1930: 77; Colonnelli, 1984: 205	Himalayas: India, Nepal, Pakistan
VI.	*Indozacladus* Colonnelli, 1984: 204 Type species: *[Ceutorhynchus] theresiae* Dalla Torre, 1922	
11	*theresiae* (Dalla Torre, 1922) *Ceutorhynchus theresiae* Dalla Torre, 1922: 125; Colonnelli, 1984: 204 *Ceutorhynchus asperulus* Faust, 1898: 323 not *Ceuthorhynchus asperulus* Boheman, 1845; Colonnelli, 2004: 66 *Indozacladus cajani* Colonnelli, 1998: 140; Colonnelli, 2004: 66	South India: India

B. Mecysmoderini Wagner, 1938a

VII. *Mecysmoderes* Schoenherr, 1837: 596
Type species: *Mecysmoderes euglyptus* Gyllenhal, 1837

12	*aurosquamosus* Korotyaev, 1989: 145	Karnataka: India
13	*carinatus* Faust, 1898: 324	Karnataka: India
14	*crassirostris* Hustache, 1924: 59	Sikkim: India
15	*notobrevicarinatus* Pajni & Kohli, 1982: 351	Northeast India
16	*notoexcavatus* Pajni & Kohli, 1982: 364	Northeast India

VIII. *Belonnotus* Schultze, 1899: 187
Type species: *Belonnotus tricarinatus* Schultze, 1899

17	*lineolatus* (Hustache, 1920) *Mecysmoderes lineolatus* Hustache, 1920: 329; Colonnelli, 2004: 72	India
18	*longicallus* (Pajni & Kohli, 1982) *Mecysmoderes longicallus* Pajni & Kohli, 1982: 345; Colonnelli, 2004: 72	Northeast India
19	*mussooriensis* (Pajni & Kohli, 1982) *Mecysmoderes mussooriensis* Pajni & Kohli, 1982: 349; Colonnelli, 2004: 72	Himalayas: North India, Nepal
20	*nigriclava* Colonnelli, 1992: 404	Uttar Pradesh: India
21	*ochraceus* (Motschulsky, 1858) *Coeliosomus ochraceus* Motschulsky, 1858: 70; Colonnelli, 2004: 72	India
22	*ochrasuturalis* (Pajni & Kohli, 1982) *Mecysmoderes ochrasuturalis* Pajni & Kohli, 1982: 360; Colonnelli, 2004: 72	Himachal Pradesh: India
23	*pectinipes* (Marshall, 1917) *Mecysmoderes pectinipes* Marshall, 1917: 406; Colonnelli, 2004: 72	Tamil Nadu: India
24	*riedeli* Colonnelli, 1992: 406	Tamil Nadu: India
25	*tenuirostris* (Marshall, 1917) *Mecysmoderes tenuirostris* Marshall, 1917: 403; Colonnelli, 2004: 72	
26	*tricarinatus* Schultze, 1899: 187	Tamil Nadu; Northeast India: India

IX. *Cysmemoderes* Colonnelli, 1992: 407
Type species: *Mecysmoderes tuberculatus* Pajni & Kohli, 1982

27 *tuberculatus* (Pajni & Kohli, 1982)

	Mecysmoderes tuberculatus Pajni & Kohli, 1982: 342; Colonnelli, 2004: 72	Manipur: India
28	*verrucosus* (Marshall, 1917) *Mecysmoderes verrucosus* Marshall, 1917: 397; Colonnelli, 2004: 72	Assam: India
X.	*Coeliosomus* Motschulsky, 1858: 70 Type species: *Coeliosomus nigrorufus* Motschulsky, 1858	
29	*brancuccii* Colonnelli, 1992: 400	Northeast India: India
30	*colonnelli* (Pajni & Kohli, 1982) *Mecysmoderes colonnelli* Pajni & Kohli, 1982: 367; Colonnelli, 2004: 72	Northeast India: India
31	*memecylonis* (Marshall, 1917) *Mecysmoderes memecylonis* Marshall, 1917: 395; Colonnelli, 2004: 72	India, Bangladesh
32	*metasternalis* (Marshall, 1917) *Mecysmoderes metasternalis* Marshall, 1917: 398; Colonnelli, 2004: 72	Himalayas: Northeast India, Myanmar
33	*minutus* (Hustache, 1924) *Mecysmoderes minutus* Hustache, 1924: 61; Colonnelli, 2004: 72	Sikkim: India
34	*neominutus* (Pajni & Kohli, 1982) *Mecysmoderes neominutus* Pajni & Kohli, 1982: 366; Colonnelli, 2004: 72	Northeast India
35	*nigrus* (Pajni & Kohli, 1982) *Mecysmoderes nigrus* Pajni & Kohli, 1982:352; Colonnelli, 2004: 72 *Mecysmoderes darjeelingensis* Pajni & Kohli, 1982: 354; Colonnelli, 2004: 72	Himalayas: North India, Nepal
36	*subhumeralis* (Marshall, 1917) *Mecysmoderes subhumeralis* Marshall, 1917: 400; Colonnelli, 2004: 73	Assam: India
37	*suturalis* (Hustache, 1924) *Mecysmoderes suturalis* Hustache, 1924: 60; Colonnelli, 2004: 73	North India
XI.	*Xenysmoderes* Colonnelli, 1992: 412 Type species: *Mecysmoderes longirostris* Hustache, 1920	
38	*alternatus* (Pajni & Kohli, 1982) *Mecysmoderes alternatus* Pajni & Kohli, 1982: 358; Colonnelli, 2004: 73	Northeast India

39 *armirufus* (Marshall, 1948)
Mecysmoderes armirufus Marshall, 1948: 455; Colonnelli, 2004: 73 — Himalayas: Northeast India, Myanmar

40 *brevicornis* (Hustache, 1924)
Mecysmoderes brevicornis Hustache, 1924: 58; Colonnelli, 2004: 73
Mecysmoderes albocapillus Pajni & Kohli, 1982: 359; Colonnelli, 2004: 73 — Himalayas: North India, Nepal

41 *comes* Colonnelli, 1992
Xenysmoderes comes Colonnelli, 1992: 413 — Himalayas: North India, Nepal

42 *humeralis* (Hustache, 1924)
Mecysmoderes humeralis Hustache, 1924: 57; Colonnelli, 2004: 73 — Himalayas: Northeast India, Nepal, Bhutan

43 *stylicornis* (Marshall, 1934)
Mecysmoderes stylicornis Marshall, 1934: 51; Colonnelli, 2004: 73 — Himalayas: North India, Myanmar, Australia

C. Egriini Pajini & Kohli, 1982

Cyphosenus subg. *Cyphosenus* Schultze, 1899: 188
Type species: *Cyphosenus paradoxus* Schultze, 1899

44 *paradoxus* Schultze, 1899
Cyphosenus paradoxus Schultze, 1899: 188 — North India

D. Hypurini Schultze, 1902

Hypurus Rey, 1882: 189
Type species: *Ceutorhynchus bertrandi* Perris, 1852

45 *portulacae* (Marshall, 1916)
Ceuthorrhynchus portulacae Marshall, 1916: 368; Colonnelli, 2004: 80 — India, Pakistan

Table 1. Number of species described during different periods

Serial Number	Period		Number of species described
	From	To	
1	1837	1900	6
2	1901	1950	18
3	1951	2000	21

Table 2. Contribution of coleopterists to Indian Ceutorhynchinae

Serial Number	Contributing coleopterists	Number of species described
1	Colonnelli	9
2	Dalla Torre	1
3	Faust	1
4	Gyllenhal	1
5	Hustache	8
6	Korotyaev	2
7	Marshall	9
8	Motschulsky	1
9	Pajni & Kohli	10
10	Schultze	3

ACKNOWLEDGEMENT

I greatly acknowledge the financial help by Kerala State Council for Science Technology and Environment under the project "Taxonomic Revision of Ceutorhynchinae (Coleoptera: Curculionidae) from India".

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(Received 06 December 2014; accepted 20 April 2015)

AUTHOR INDEX

Statement of ownership and other particulars of ENTOMON

(Form IV, Rule 8 of Registration of Newspapers (Central) Rules 1956)

1.	Place of publication	:	Trivandrum
2.	Periodicity of publication		Quarterly
3.	Printer's name, nationality and address	:	Dr K D Prathapan, Indian, Secretary, Association for Advancement of Entomology, Department of Entomology, College of Agriculture, Kerala Agricultural University, Vellayani PO, Thiruvananthapuram 695522, Kerala, India
4.	Publisher's name, nationality and address	:	- do-
5.	Editor's name, nationality and address	:	Dr M S Palaniswami, Indian, Chief Editor, ENTOMON, Association for Advancement of Entomology, Department of Entomology, College of Agriculture, Kerala Agricultural University, Vellayani PO, Thiruvananthapuram 695522, Kerala, India
6.	Name and address of the Individual who owns the paper	:	Association for Advancement of Entomology, Department of Entomology, College of Agriculture, Kerala Agricultural University, Vellayani PO, Thiruvananthapuram 695522, Kerala, India

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Dr K. D. Prathapan
Publisher, ENTOMON

Vellayani PO, Thiruvananthapuram 695522
30 May 2015

Reg. No. 123SBN

Layout and printing at SB Press (P) Ltd, Trivandrum-1